## APPENDIX

## VERSION WITH MARKINGS TO SHOW CHANGES MADE

1. (Amended) [A simple and accurate method for assay of a single-stranded RNA containing a specific nucleic acids sequence in a sample at almost constant temperature by using at least the following reagents (A) to (I), which comprises a step of adding the reagents (A) to (I) one by one (in any order), in combinations of at least two or all at once and a step measuring a fluorescent signal in the presence of the reagent (I) at least once after addition of at least the reagents (A) to (H);

(A) a first single-stranded oligonucleic acid complementary to a sequence neighboring the 5' end of the specific nucleic acids sequence in the single-stranded RNA,

(B) a second single-stranded oligo DNA complementary to a 3'-end sequence within the specific nucleic acids sequence,]

A method for assaying a single-stranded RNA in a sample, wherein said RNA contains a specific nucleic acid sequence, said method comprising the following steps:

(1) adding to a reaction vessel containing said single-stranded RNA, reagents (A) to (J), wherein said reagents are added to the reaction vessel at constant temperature and one by one, or in combinations of at least two, or all at once: and

(2) after addition of reagents (A) to (I), measuring, at least once, a fluorescent signal in the presence of reagent (J); and

wherein said reagents (A) to (J) are as follows,

(A) a first single-stranded oligo nucleic acid complementary to a sequence 5' of, and adjacent to, the 5' end of said specific nucleic acid sequence,

(B) a second single-stranded oligo DNA complementary to a sequence at the 3' end of said specific nucleic acid sequence,

(C) an RNA-dependent DNA polymerase,

(D) a ribonuclease that degrades RNA in a DNA-RNA double-strand,

(E)[(D)] deoxyribonucleoside triphosphates,

(F)[(E)] [a third single-stranded oligo DNA having (1) a promoter sequence for a DNA-dependent RNA polymerase, (2) and enhancer sequence for the promoter and (3) a 5 '-end sequence within the specific nucleic acids sequence, in this order from the 5' end,] a third single-stranded oligo DNA having at the 5' end of said oligo DNA the following sequences, in the following order, proceeding in a 5' to 3' direction with respect to the third single-stranded oligo DNA: 1) a promoter sequence for a DNA-dependent RNA polymerase, 2) an enhancer sequence for said promoter, and 3) a sequence at the 5' end of said specific nucleic acid sequence,

(G)[(F)] a DNA-dependent DNA polymerase,

(H)[(G)] a DNA-dependent RNA polymerase,

(I)[(H)] ribonucleoside triphosphates, and

(J)[(I)] a fourth single-stranded oligo DNA complementary to [the] said specific nucleic acid[s] sequence [which], wherein said fourth single-stranded oligo DNA is labeled so that it gives off a measurable fluorescent signal upon hybridization with a nucleic acid containing [the] said specific nucleic acid[s] sequence.

2. (Amended) The method according to Claim 1, wherein the temperature is selected from the range of from 35 to 60°C.

3. (Amended) The method according to Claim 1, wherein the first [oligonucleic] oligo nucleic acid as the reagent (A) is a DNA, and the method further comprises a step of adding an RNaseH and a subsequent step of deactivating the RNaseH by heating or by addition of an inhibitor prior to addition of the reagent (B).

4. (Amended) The method according to Claim 3, wherein addition of the reagent (A) is followed by simultaneous addition of the reagents (B) to [(H)](I), and further by addition of the reagent [(I)](J).

5. (Amended) The method according to Claim [(3)]3, wherein addition of the reagent (A) is followed by simultaneous addition of the reagents (B) to [(I)](J).

6. (Amended) The method according to Claim 1, wherein the first [oligonucleic] oligo nucleic acid as the reagent (A) is a ribozyme or a DNAzyme.

Please cancel claims 7 and 8.

9. (Amended) The method according to Claim [7]1, wherein the enzyme which degrades RNA in a DNA-RNA double strand is the RNA-dependent DNA polymerase as the reagent [(C)](D).

10. (Amended) The method according to Claim 1, wherein an enzyme having both an RNA-dependent DNA polymerase activity and a DNA-dependent DNA polymerase activity is used as the reagents (C) and [(F)](G) to [virtually]essentially omit addition of the reagent (C) or the reagent [(F)](G).

11. (Amended) The method according to Claim 10, wherein the enzyme is avian [myoblastome] myoblastoma virus polymerase.

12. (Amended) The method according to Claim 1, wherein the second and third oligo DNAs as the reagents (B) and [(E)](F) are used at concentrations of from 0.02 to 1 μM.

13. (Amended) The method according to Claim 1, wherein the DNA-dependent RNA polymerase as the reagent [(G)](H) is at least one enzyme selected from the group consisting of phage SP6 polymerase, phage T3 polymerase, and [phase] phage T7 polymerase.

14. (Amended) The method according to Claim 1, wherein the fourth oligo DNA as the reagent [(I)](J) is a DNA which is linked to a fluorescent intercalative dye so that the fluorescent intercalative dye changes its fluorescence characteristic upon hybridization of the DNA with another nucleic acid by intercalating into the resulting double strand.

15. (Amended) The method according to Claim 1 or 14, wherein the fourth oligo DNA as the reagent [(I)](J) is a DNA which has a 3'end sequence [uncomplimentary] that is not

complementary to the specific nucleic acid[s] sequence or has a modified 3' end, and hybridizes to the nucleic acid of Claim 1 having said specific nucleic acid sequence.

16. (Amended) The method according to Claim 1, which further comprises a step of detecting or quantifying the single-stranded RNA in the sample based on the measured fluorescent signal or change in the measured fluorescent signal.

17. The method according to Claim 1, wherein all the reagents are chloride-free.

18. (Amended) The method according to Claim 1, [which further uses] wherein step (2) further comprises the addition of an acetate.

19. The method according to Claim 18, wherein the acetate is magnesium acetate at a concentration of from 5 to 20 mM or potassium acetate at a concentration of from 50 to 200 mM.

20. (Amended) The method according to Claim 1, [which further uses] wherein step (2) further comprises the addition of sorbitol.

Please cancel claims 21, 22, and 23.

Please add new claim 29 as follows:

--29. The method according to claim 1, wherein the first oligo nucleic acid as the reagent (A) is a DNA.--